replication. This increased mutation rate, in turn, can promote the development of drug resistance under stress.
Here we employ structural modeling, saturation mutagenesis, and kinetics to systematically elucidate substrate preference of the LexA enzyme in Pseudomonas aeruginosa. Our studies elucidate the key determinants for substrate recognition and demonstrate that overall, LexA is a relatively tolerant protease. Furthermore, this study reveals positions in the LexA protein that are amenable for labeling with fluorescent probes, which can be used for conformational studies and inhibitor screens. Understanding the enzymes that regulate mutation can lead to insights about the evolution of bacteria and potentially open up new avenues for combating the rise of drug resistance

1193-Pos Board B85
An Adaptation of Prostaglandin Endoperoxide H Synthases in Binding to Endogenous Ligands and Nonsteroidal Anti-Inflammatory Drugs
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Prostaglandins play critical roles in a number of physiological and pathophysiological processes. These molecules are involved in regulating blood flow to organs, initiating platelet aggregation in blood clotting, and mainly mediating the classical symptoms of inflammation. Biosynthesis of prostaglandins is initiated by prostaglandin endoperoxide H synthases (PGHS) −1 and −2, which catalyze the conversion of arachidonic acid to PGH_2. PGHSs are homodimers comprised of ~72 kDa monomers of identical primary sequence and recently have been investigated in the respect of allosteric communications between two subunits during ligand binding. The nonsteroidal anti-inflammatory drugs show multiple inhibition modalities against PGHSs. Their time-dependent and subunit-preferential bindings to PGHSs complicate the interactions of PGHSs with the cognate substrates and their regulations by natural and synthetic ligands. Accordingly, a paradigm shift in the analysis of PGHS-ligand complex during PGHS catalysis is required and their theoretical revaluation and biological functioning *in vivo* are described in this paper.

1194-Pos Board B86
Biochemical Aspects of Characterization of Human Selenoprotein M and its Putative Role in Oxidative Stress
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The biomedical functions of essential trace element selenium have been attributed to selenoproteins, a class of proteins containing genetically encoded 21st amino acid selenocysteine (single letter code U). Besides thioredoxin reductase and glutathione peroxidases, two well-studied examples of the selenoproteome family, the enzymatic functions of other family members remain elusive. About one third of these selenoproteins possess a minimal thioredoxin fold with the reactive selenocysteine is positioned on a flexible loop where it is easily accessible to substrates. This study focuses on characterization of a representative member of the family - the human selenoprotein M (SelM). SelM is localized to endoplasmic reticulum and is highly expressed in brain where it protects the neurons from oxidative stress. We report that SelM is a peroxidase and that its enzymatic activity depends on the presence of selenocysteine. In addition, we discuss the oligomerization state of the protein and approaches to map the redox potential of SelM and other members of the family. Since the redox potential of selenoproteins is by large unknown, the determination of this redox potential of selenoproteins with minimal thioredoxin fold will help us understand their physiological functions.

1195-Pos Board B87
In the Yeast Saccharomyces Cerevisiae Actin Stabilizes a Glycolytic Pathway Enzyme Complex
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Metabolic pathway enzymes may associate into supramolecular complexes increasing efficiency and protecting each other from inhibitors and physical stress. The eukaryotic cytoplasm is a concentrated, highly organized mesh of fibers, macromolecules and solutes. Here, the associations of specific proteins would result in intermediary channeling. In addition, enzymes in complexes are probably protected against inhibition during the stress response, when compatible solutes rise. However, detection of complexes is difficult due to the weak nature of enzyme associations; this difficulty was circumvented by adding F-actin in order to stabilize association and further modify activity and stability of the complexed enzymes. Polymeric F-actin, but not the monomeric G-actin stabilized glycolytic enzyme complexes and promoted protection against inhibition by either trehalose or by specific anti-enzyme antibodies. The association of specific enzymes and even the whole fermentation pathway were stabilized by F-actin, producing a functional pathway that resisted inhibition by either trehalose or by antibodies.

1196-Pos Board B88
Mutational Analysis of the Integral Membrane Methyltransferase ICMT
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The eukaryotic enzyme isoprenylcysteine carboxylmethyltransferase (ICMT) catalyzes the attachment of a methyl group onto the carboxylate of a lipid-modified cysteine residue at the C-terminus of its protein substrates. This is the final processing step for proteins that contain a C-terminal 'CAAX' motif, including Ras. Because inhibition of ICMT blocks Ras-induced oncogenic transformation, ICMT is a target for cancer therapy. Human ICMT is predicted to have eight transmembrane helices and it has no discernable homology with soluble methyltransferases. A salient question in understanding the mechanism of this enzyme is how both its water-soluble substrate (S-adenosyl-L-methionine, AdoMet) and its lipophilic substrate (a prenylated cysteine residue) are recognized in the active site. A crystal structure of an integral membrane methyltransferase from a prokaryotic organism was recently determined. The prokaryotic enzyme, which contains five transmembrane helices, has sequence conservation with ICMT in the region that binds AdoMet but has limited conservation elsewhere. In order to identify regions of ICMT that are important for catalytic activity, we mutated roughly half of the residues in ICMT and we interpreted the results with regard to the available structure. The mutants with reduced activity can be classified into two groups. The first class contains residues that form the AdoMet binding site and are conserved with the prokaryotic enzyme. For the second class of inactivating mutants, which is more numerous, there is high sequence conservation among eukaryotic ICMT homologs, but no discernable sequence conservation with the prokaryotic enzyme. This second class may contain residues involved in recognizing the prenylated cysteine substrate in eukaryotes. Our results indicate that the structure of the prokaryotic enzyme is informative for understanding the AdoMet binding site of ICMT and they point to differences between these two enzymes in other respects.

Intrinsically Disordered Proteins II

1197-Pos Board B89
Modulation of Polyglutamine Conformations and Associations by C-Terminal Proline Rich Regions from Exon 1 of Huntingtin
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Huntington's disease (HD) is associated with CAG repeats within exon 1 that lead to polyglutamine (polyQ) expansions in the protein huntingtin (htt). Proteolysis of htt leads to products that can be trafficked into the nucleus and form neuronal intranuclear inclusions. The presence / absence of flanking sequences namely, the N-terminal 17-residue amphipathic stretch (Nt17h-htt) and the C-terminal 38-residue proline-rich stretch (Ct38h-htt) in products of proteolysis have a profound effect on the HD phenotype in animal models (where h-htt denotes human-htt). The biophysical basis for the effects of these flanking sequences remains unresolved. Here, we focus on the influence of different proline-rich C-terminal sequences on the conformations of and intermolecular associations through the polyQ tract. The flanking sequences include Ct38h-htt and Ct31m-htt (the mouse 31-residue C-terminal proline-rich region).
We used atomistic simulations based on the ABSINTH implicit solvation model and underlying forcefield paradigm to quantify the nature of the coupling between C-terminal flanking sequences and polyQ tracts for different polyQ lengths. There are clear differences between the intrinsic and context dependent properties of Ct31m-htt and Ct38h-htt. Detailed analyses of the monomer conformations and intermolecular associations of polyQ in cis with different synthetic and naturally occurring C-terminal proline-rich stretches identify how flanking sequences modulate aggregation mechanisms. These results might be important for recent studies in animal models that show different toxicity profiles between transgenic animals expressing different C-terminal proline-rich sequences.
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1198-Pos Board B90
Measuring Disorder and Dynamical Properties of FG-Nucleoporins
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The nuclear pore complex (NPC) is a massive protein structure located on the nuclear envelope serving as a selective gateway between the cellular